

CLAIMS

That which is claimed is:

- 5 1. A method to determine the optimal length of an immunobiologically-active liner peptide epitope of a polypeptide, the method comprising the steps of:
 - a) providing a curve characterizing the hydrophilicity and/or hydrophobicity of the linear sequence of amino acid residues of a polypeptide;
 - b) generating at least one potential epitope set comprising at least one potential
10 epitope by fitting a window of the curve of step (a) to a mathematically generated continuous curve, the continuous curve having repeating values at regular intervals with at least a maximum positive value, the window containing a specific number of amino acid residues and the window is lagged through the curve of step (a);
 - c) increasing the number of residues in the window after each lagging;
 - 15 d) determining and ranking potential epitopes for each set by selecting potential epitopes having a positive-fit correlation value determined by fitting curves in step (b) thereby providing a set of ranked potential epitopes for each window of residues used in step (b), the most positive-fit correlation value ranked first in each potential epitope set;
 - e) examining the positioning of at least the highest ranked potential epitopes of each
20 set relative to the linear sequence of the plot of step (a) to determine at least one set of potential epitopes that exhibit alternating positioning about an equilibrium position wherein the ranking values of the potential epitopes converge towards or diverge away from the equilibrium position; and
 - f) designating the potential epitopes of the set having the most alternating ranking
25 values that converge or diverge as the immunologically active epitopes which have an optimal length equating to numeric value of amino acid residues in the potential epitopes.
2. The method according to claim 1 wherein the mathematically generated curve is generated by a negative cosine curve function.

3. A method to determine the optimal length of an immunobiologically active linear peptide epitope of a polypeptide characterized by a hydrophobic-hydrophilic-hydrophobic motif, the method comprising the steps of:

- 5 a) assigning an average hydropathy value to each amino acid of the polypeptide;
- b) generating a hydrophilicity plot using the average hydropathy value of each amino acid;
- c) fitting a curve segment of the hydrophilicity plot to a negative cosine function, wherein a specific period number value of the negative cosine function equates to the number
10 of amino acids in the curve segment, the period number increasing within a predetermined chosen period number range after each sequential lagging through the hydrophilicity plot thereby providing fit-correlation values for each curve segment across the linear sequence when using the specific period number value;
- d) generating a potential Ho-Hi-Ho epitope set for each specific period number
15 value within the chosen period number range, wherein each potential Ho-Hi-Ho epitope set contains potential Ho-Hi-Ho epitopes that have a fit- correlation value;
- e) ranking each potential Ho-Hi-Ho epitope in the potential Ho-Hi-Ho epitope set according to positive fit-correlation values wherein the epitope having highest positive-fit correlation value is ranked number one thereby providing ranked Ho-Hi-Ho potential epitopes
20 for each specific period number value;
- f) examining the positioning of at least the highest ranked Ho-Hi-Ho potential epitopes of each set relative to the linear sequence of the plot of step (a) to determine at least one set of Ho-Hi-Ho potential epitopes that exhibit alternating positioning about an equilibrium position wherein the ranking values of the Ho-Hi-Ho potential epitopes converge towards or
25 diverge away from the equilibrium position; and
- g) designating the Ho-Hi-Ho potential epitopes of the set having the most alternating ranking values that converge or diverge as the immunologically active epitopes which have an optimal length equating to numeric value of amino acid residues in the potential epitopes.

4. The method according to claim 3 wherein said hydrophilicity curve is generated using Kyte-Doolittle hydropathy values with reversed signs.
5. The method according to claim 3 further comprising choosing the potential epitope set having the highest fit correlation value found in step (c) if more than one potential epitope set exhibits the same number of alternating ranking values as examined in step (f).
6. A Ho-Hi-Ho epitope of a polypeptide, said Ho-Hi-Ho epitope characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues determined by method of claim 3.
7. The Ho-Hi-Ho epitope according to claim 6 wherein the amino acid residues are altered by replacing amino acids to increase or decrease the fit correlation between the hydrophilicity curve and the negative cosine curve thereby increasing or decreasing the affinity for the epitope by immune components.
8. A method for determining the viability of a protein comprising:
- a) finding the immunobiologically active epitopes of a polypeptide and their optimal length according to the methods of claim 3; and
 - b) comparing the optimal length found in step (a) to the optimal length found in anti-polypeptide antisera.
9. A antisera specific for a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is defined by a motif of two hydrophobic and one hydrophilic regions arranged in the following manner

hydrophobic - hydrophilic - hydrophobic

wherein said epitope has an optimal length of amino acid residues determined by method of claim

3.

10. An antigenic composition comprising a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues determined by method of claim 3.

11. The antigenic composition comprising a nucleic acid molecule coding for a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues determined by method of claim 3.

12. A diagnostic testing method comprising the steps of:

- (i) providing a sample
- (ii) contacting said sample with antisera specific for a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues determined by method of claim 3; and
- (iii) detecting the binding said antisera to a polypeptide in said sample.

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13. A method to determine the optimal length of an immunobiologically-active linear peptide epitope of a polypeptide, the method comprising the steps of:

- a) providing a hydrophilicity and/or hydrophobicity plot generated for the amino acid linear sequence of a polypeptide ;
- b) fitting the plot of step (a) to a mathematically generated continuous curve thereby generating potential epitope sets which include ranked potential epitopes having a specific number of amino acid residues; and
- c) comparing the sets of ranked potential epitopes to other generated data to determine the immunobiologically-active linear peptide epitope and its optimal

length.

14. The method according to claim 13 wherein the other generated data of step (c) is selected from the group consisting of: comparing magnitude of oscillating behavior, comparing
5 the ranked potential epitopes with other epitopes generated by propensity scales, comparing with a previously generated plot and combinations thereof.

~~15.~~ A method to determine the optimal length of an immunobiologically-active linear peptide epitope of a polypeptide, the method comprising the steps of:

- 10 a) fitting a hydrophilicity and/or hydrophobicity plot generated for the amino acid linear sequence of a polypeptide to a mathematically generated continuous curve thereby generating potential epitope sets which include ranked potential epitopes having a specific number of amino acid residues, the mathematically generated curve having at least a maximum positive value;
- 15 b) positioning the ranked potential epitopes for each set on the hydrophilicity and/or hydrophobicity plot to determine the oscillating behavior of the numeric value of ranked potential epitopes; and
- c) deeming the potential epitopes that exhibit the most alternating positioning about an equilibrium position when juxtaposed on the hydrophilicity and/or
20 hydrophobicity plot as the theoretical epitopes and their optimal length corresponds to the specific number of amino acid residues in the set of ranked potential epitopes.

16. The method according to claim 15 wherein said hydrophilicity curve is generated using
25 Kyte-Doolittle hydropathy values and the mathematically generated curve is generated by a negative cosine function having a period number equivalent to the window of residues.

17. A Ho-Hi-Ho epitope of a polypeptide, said Ho-Hi-Ho epitope characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues

determined by method of claim 16.

18. A antisera specific for a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is defined by a motif of two hydrophobic and one hydrophilic regions arranged in the following manner

hydrophobic - hydrophilic - hydrophobic

- wherein said epitope has an optimal length of amino acid residues determined by method of claim 15.

19.

A diagnostic testing method comprising the steps of:

- (i) providing a sample
- (ii) contacting said sample with antisera specific for a Ho-Hi-Ho epitope of contiguous amino acid residues from a polypeptide wherein said epitope is characterized by a hydrophobic-hydrophilic-hydrophobic motif having an optimal length of amino acid residues determined by method of claim 15; and
- (iii) detecting the binding said antisera to a polypeptide in said sample.

20. A antigenic composition comprising a nucleic acid molecule coding for an epitope of contiguous amino acid residues from a polypeptide wherein said epitope has an optimal length of amino acid residues determined by method of claim 15.